1 gatgatgaca ctattgctaa ggtcaacgct gcaatgaatg gacagttgcc

51 gtttgctgtt gtgggaagta tggatgaggt aaaagtcgga aacaagatgg

101 tcaaagctcg ccagtaccct tggggtgttgt acaagtggaa aatgaaaacc

151 actgtgactt tgtaaagctg cgggaaatgc tcatttgtac aaatatggag

201 gacctgcgag agcagaccca taccaggcac tatgagcttt acaggcgctg

251 caaactggag gaaatgggct ttacagatgt gggcccagaa aacaagccag

301 tcaggtaggg tggggttctt gggaagccaa ccaaagagga agttagagag

351 gattgcatat ttttatatat ccaggatgat ggggcatgtg ttctttagaa

401 tctcccaaaa ccatgatata gaatcatttg gtaatgacta attattgtgc

451 ttctt

Figure 1. Partial cDNA sequence of Shinc-1 gene.
The partial nucleotide sequence of a Shinc-1 cDNA fragment (456 bp) isolated from human prostate cancer cells (DU-145) by the differential display of mRNA approach is shown.

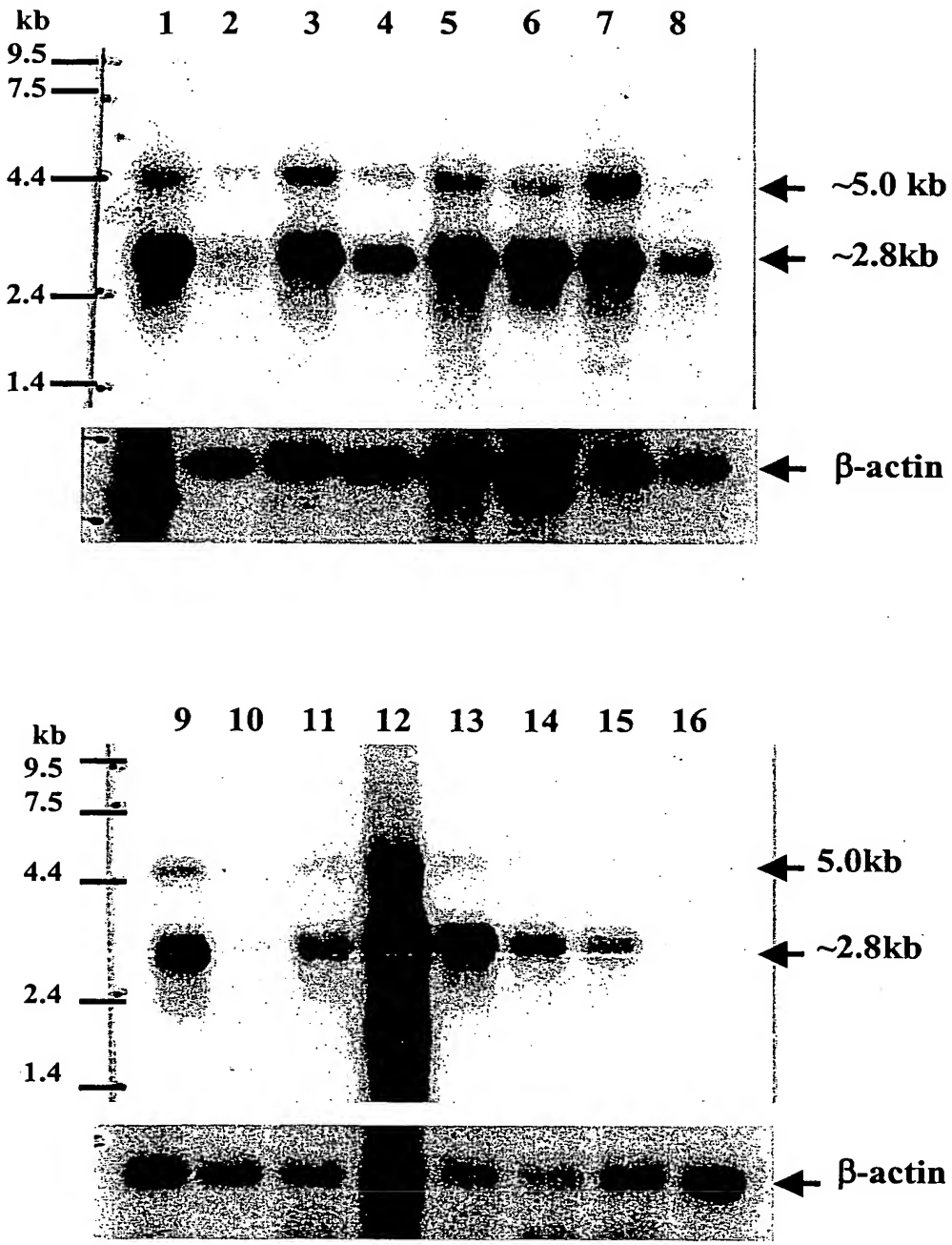


Figure 2. Expression on Shinc-1 mRNA in normal human tissues.
Blots were hybridized with radiolabeled Shinc-1 cDNA probe followed by β-actin cDNA probe.
Lane 1: Heart; lane 2: Brain; lane 3: Placenta; lane 4: Lung; lane 5: Liver; lane 6: Skeltal muscle; lane 7: Kidney; lane 8: Pancreas;
lane 9: Spleen; lane 10: Thymus; lane 11: Prostate; lane 12: Testis; lane 13: Ovary; lane 14: Small intestine; lane 15: Colon; lane 16: Peripheral blood leukocytes. Approximately 5.0 kb and 2.8 kB Shinc-1 transcripts are shown.

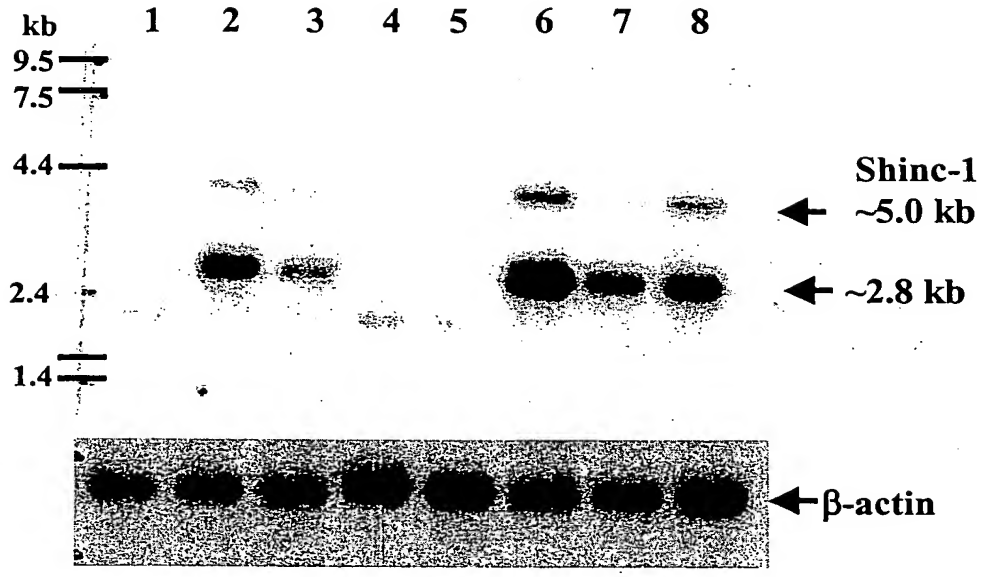
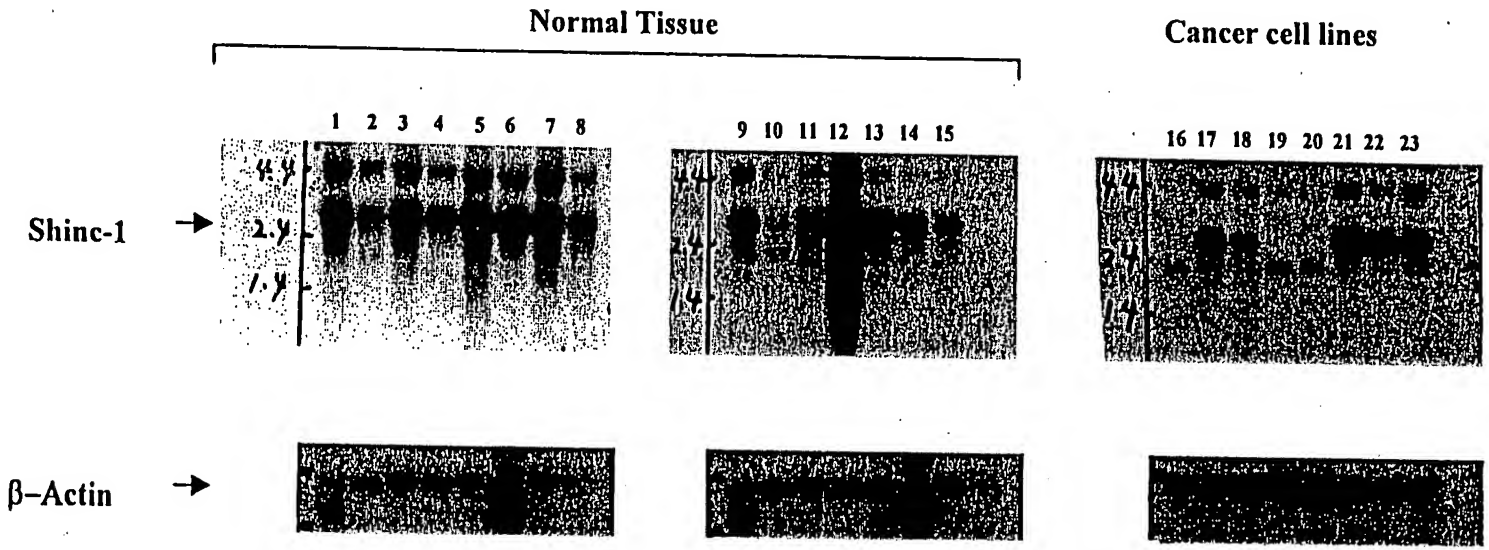


Figure 3. Expression of Shinc-1 mRNA in human cancer cells.
Blots were sequentially probed with radiolabeled Shinc-1 (upper panels) and β-actin (lower panels) cDNA probes. HL-60, promyelocytic leukemia (lane 1);HeLa-S3, (lane 2); K-562, chronic myelogenous leukemia (lane 3); MOLT-4, lymphoblastic leukemia (lane 4); BL-RAJI, Burkitt's lymphoma (lane 5); SW480, colorectal adenocarcinoma (lane 6); A549, lung carcinoma (lane 7); G361,melanoma (lane 8).

FIGURE 4


Normal Tissue
Cancer cell lines
1 2 3 4 5 6 7 8
9 10 11 12 13 14 15
16 17 18 19 20 21 22 23
4.4
2.4
1.4
Shinc-1
β–Actin

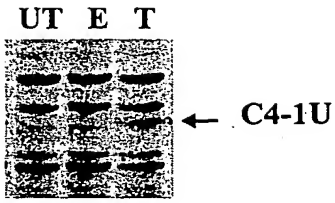


Figure 5. Identification of differentially expressed C4-1U cDNA fragment in DU-145 cells treated with tempo. DU-145 cells were treated for 2 h with 7.5 mM tempo (T), or vehicle (ethanol 1%. E) or left untreated (UT). Total cellular RNA was extracted using RNAzol B (Tel-Test Inc, Texas).

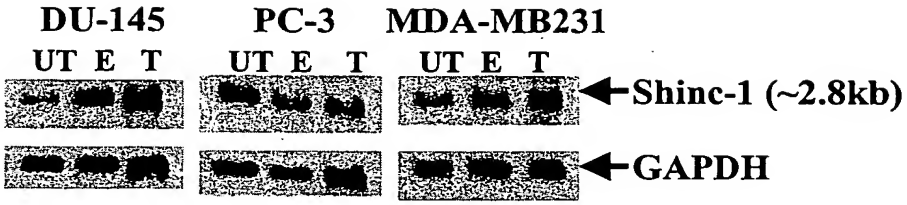


Figure 6. Northern blot hybridization analysis of Shinc-1 gene expression in tempo-treated human prostate (DU-145 and PC-3) and breast cancer cells (MDA-MB 231).

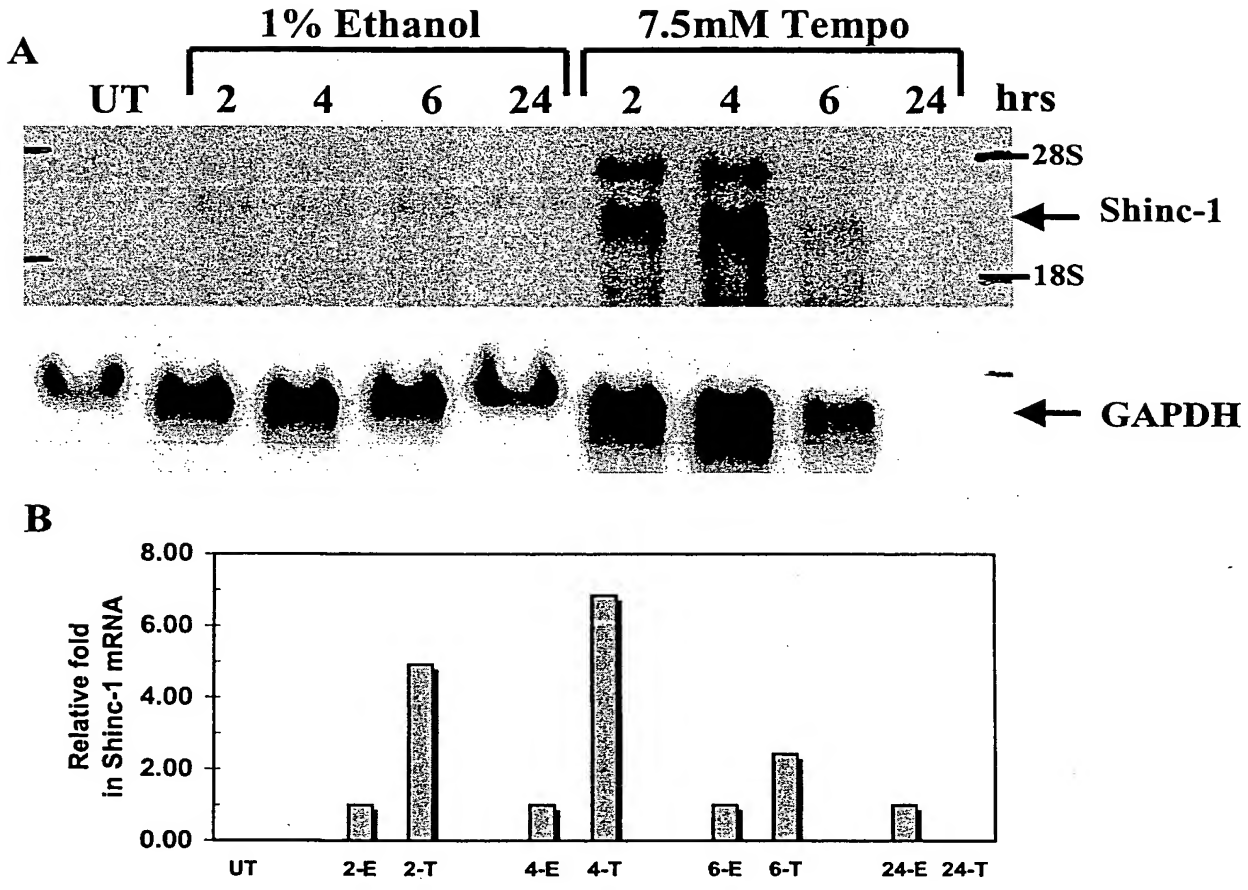


Figure 7. Time-course analysis of Shinc-1 mRNA expression in tempo-treated cells
(A) DU-145 cells were treated with 7.5mM tempo or 1% ethanol for the indicated time.Total RNA were extracted from the cells and fractionated by electrophoresis.
(B) Autoradiographs were computer-scanned using the Image-Quant software (Molecular Dynamics). Relative fold change in the steady-state mRNAs level were calculated by normalizing against the GAPDH signal, followed by comparison with the expression in ethanol-treated and tempo-treated cells. UT: untreated, E: ethanol, T: tempo